DETERMINATION OF FAECAL POLLUTION IN THE KUBANNI LAKE AND RIVER USING FAEGAL COLIFORMS AND FAECAL STREPTOCOCCI AS INDICATORS

BY

Charles Okuofu (B.Sc)

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THESIS APPROVAL

I hereby recommend that the thesis prepared under my supervision by Mr. Christian Okonfo (B.Sc.) entitled:

"Determination of Faecal Pollution In the Kubanni Lake and Stream Using Faecal Coliform and Faecal Streptococci as indicators", be accepted in partial fulfilment of the requirements for the degree of MASTER OF SCIENCE.

DR. J. B. GAULTNEY
(Principal Supervisor)

DR. J. A. OGUNKOMBI
(CO-Supervisor)

PROF. NDUKA OKAFOR
(External Examiner)

DR. (MRS.) A. M. POKRZYWA
(CO-Supervisor)

DATE: July 10, 1978

C O N T E N T S

Title Page

Thesis Approval Page

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D E D I C A T I O N

You tended and cared

For me till date

You took pains and suffered

For me till date

You worried and bothered

For me till date

Sweet mother!

This is for you with love.

Charles A. Okuofu.

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S U M M A R Y

The effectiveness of the sewage treatment plant at A.B.U. Zaria in reducing bacterial numbers was examined and the potability of water in Kubanni Lake, also at A.B.U., was determined. Samples of water and sewage were taken once a week for 41 weeks and analysed. Sampling sites were distributed so that samples of raw sewage, of effluent and of lake water were tested throughout this study. Some samples also were taken down-stream from the entrance of the effluent into the Kubanni River.

Faecal coliform and faecal streptococcal densities were determined by the surface plate (direct count) method, using EMB agar for faecal coliform isolation and M-enterococcus agar for faecal streptococci. The densities of these organisms are compared and discussed.

The sewage treatment plant at A.B.U. which is an oxigest package plant model R, is overloaded and is not functioning properly. Reduction in bacterial numbers between raw sewage and effluents is minimal. Mean faecal bacteria counts were 1.4×10^7/100ml faecal coliforms and 3.2×10^7/100ml faecal streptococci at the outlet from the treatment plant. A high BOD in the effluent was such that the faecal indicator bacteria were able to maintain a steady population.

Faecal coliform density at point of effluent discharge into Kubanni River was in the order of 1.9×10^7/100ml while faecal streptococcal density was 4.9×10^6/100ml. These levels of faecal bacteria

constitute a public health hazard to users of the water below the A.B.U. sewage treatment plant. Counts were still 2.3×10^3/100ml faecal coliforms and 1.3×10^3/100ml faecal streptococci after flowing 600 meters donstream. These sampling sites extended past Jama'a village and into an area used for grazing of cattle. Since high counts of faecal indicator bacteria were found and 5 Salmonella Sp were isolated from effluent, the untreated effluent is not suitable for irrigation as is currently practised by the local farmers, nor is it suitable for use as drinking water for cattle.

The Kubanni Lake water was found to be unpolluted most of the time. There were 98% negative samples for faecal coliforms and 91% negative samples for faecal streptococci throughout the sampling period.

Suggestions for improvement on sewage treatment are made.

INTRODUCTION

Communities in many parts of the World are still inadequately supplied with potable water. Most people in developing countries including Nigeria depend largely on untreated ground and surface waters for drinking and other purposes. This practice is fast becoming undesirable because of the pollution problem. According to World Health Organisation (Director General, 1976), the proportion of population served by piped water supply in urban areas, rose from 50% in 1970 to 57% in 1975. By 1980, the percentage is expected to be 68%. However, in rural areas, the proportion of population having access to safe water rose from 14% to 22% in the period 1970-75. The set target for 1980 is 25%. The World Health Organisation has been striving to improve the water supplies in developing countries. To this end, the organisation has placed a number of technical services at the disposal of member states, to assist them in the study of water quality problems.

The major sources of water pollution are the domestic, municipal sewage, as well as industrial and agricultural wastewaters. The discharge of sewage, industrial and agricultural wastes into water introduces decomposable organic matter in the water. The degradation of such organic matter exerts a demand on dissolved oxygen (DO), which affects the quality of the water. Toxic substances from industries or pesticides from agricultural wastes may find their way into water bodies. The various contaminants such as bacteria,

toxic materials and heavy metals as well as long chain hydrocarbons when discharged in watersm are health hazards. They may be responsible for the waterborne diseases such as cholera and typhoid fevers; and also poisoning due to toxic chemical substances.

The various methods of wastewater treatment are aerated lagoons, stabilisation ponds, trickling filters and activated sludge systems etc. These methods are continually being improved and modified. The efficiency of these treatment methods in Biochemical Oxygen Demand (BOD) removal are as follow: Aerated lagoons 75-90%; waste stabilisation ponds 75-90%; and activated sludge process 90-93%. (Arceivalla, 1973). All these treatment methods result in reduction of both pathogenic and other bacteria numbers in wastewaters.

At Ahmadu Bello University (ABU) Zaria, the sewage treatment plant is the "Oxigest package plant mode 'R'.(see appendix E). This is a type of activated sludge treatment plant operating on contact stabilisation principles. In this process, raw sewage enters the plant at the inlet pipe and mixes with aerated sludge in the contact zone. In this chamber, aerobic decomposition by bacteria takes place. Organic matter is rapidly removed. Fresh sewage entering the plant displaces mixed liquor from the contact zone into the settling basin. The activated sludge falls to the bottom of the settling basin and is scraped into the central sludge well by mechanically driven scraper. Supernatant liquid from the central basin flows over the notched weir into the effluent trough. From here, it leaves the plant through the

outlet pipe. Grease and other floating matter in the central settlingg basin is directed mechanically by a skimmer into a scum trough and from there to the aerobic digester. Some sludge is drawn from the settling basin by an air - lift into the aeration zone. This sludge flows slowly around the aeration zone and is thoroughly aerated. When it enters the contact zone again, it is fully conditioned and ready to treat fresh sewage. About 1500cm^3 of effluents are estimated to be discharged unchlorinated, into River Kubanni daily (see fig 5). The effluents flows through a natural gully about 600m long before eventually entering the river. .

The release of effluents with high organic matter content affects the life of bacteria in the effluents. Organisms like the coliform bacteria will multply, and if the organic matter content is high enough, faecal streptococci will also proliferate in effluents (Morris and Weaver, 1954). Some of the effluents are used by the local farmers to irrigate their farms (figs. 6 & 7). Among the crops they cultivate are tomatoes, spinach, onions, sugar-cane, pepper, rice and cabbages. Apart from the danger posed by this practice to the farmers themselves, their crops are sold in the markets for consumption by the general public. This is a serious public health problem because of possible contamination of the crops.

Jama'a village is situated below the point of discharge of effluents into river Kubanni. (Fig.5). Fortunately, the villagers do not use the river as a source of water for domestic purposes. They depend on wells for these. However, children play in and around the river and cattle graze along its banks. If this river is polluted as is suspected, then a danger exists because Salmonella sp can be spread from animals to man through meat (Watson 1975). Also, the possibility of direct infection of the children playing in the water is not ruled out. It was observed that the Kubanni valley is dry during the dry season and only a little seepage of water occurs from the nearby Kubanni Lake. Therefore, most of the water flowing down the river bed during the dry season is untreated effluents from the treatment plant. Very little or no dilution (due to seepage from the Lake), occurs during this period. Consequently, there is need to keep the organic matter in A.B.U. effluents at a very low level to reduce the dangers.

Kubanni dam is located upstream and above the effluent discharges point. The reservoir created by the dam is to be used as a source of water supply for the University. It receives several small tributaries, one of which is Samaru Stream. Samaru Stream flows through Samaru village, an abattoir, A.B.U. junior staff quarters, passes near a septic tank in Dan Fodio Hall (seepage from this enters the stream); near ICSA hall and the University's incinerator. The stream carries along wastes and pollutants from all these sources, which should affect the quality of water in the lake (see fig. 1).

This research has two principal objectives. The first objective is to determine the effectiveness of the existing A.B.U. treatment plant in removing faecal coliforms and faecal streptococci from sewage, and the numbers of these bacteria in effluents and Kubanni River. The second objective is to investigate the potability of water over and around the intake structure leading from Kubanni Lake to the proposed water purifications plant at A.B.U Zaria. In addition to these, the value of faecal coliforms and faecal streptococci as indicators of water pollution will be determined.

LITERATURE REVIEW

Determination of pollution of a water is based on the recovery of pollutants from the tested water. These pollutants may be chemical or biological matter, like mercury or bacteria. The presence of bacteria in drinking water may not necessarily indicate that the water is non-potable. It is the presence of harmful chemicals or pathogenic microbes that render water non-potable. The water borne microbial organisms harmful to man are usually of intestinal origin either from man or animals. These include protozoa like E histolytica, viruses like infectious hepatitis virus and bacteria like Salmonella sp. The isolation of these pathogens from water is a difficult task, being often expensive and time consuming. Also, numbers of the pathogens in water are relatively low. Therefore, the need has arisen for organisms which can reliably

indicate the presence of pathogens. Such organisms must satisfy certain conditions.

Bondo, (1966). Summarises the conditions as follow:-

- The organism must be present whenever the pathogens are present.
- It must be present only when presence of pathogens is an imminent danger i.e. the organism should not multiply to any great extent in aqueous environments.
- Occur in much greater numbers than the pathogens
- Be more resistant to disinfection and the aqueous environment.
- Grow readily on relatively simple media
- Yield characteristic but simple reactions to enhance unambiguous identification.
- Be randomly distributed in the sample to be examined, and
- Grow widely, independent of other organisms, when inoculated in artificial media i.e. should not be inhibited.

Most of the pathogens encountered in water are not suitable indicators of pollution as they do not satisfy the above conditions. On the other hand, there are bacteria which satisfy many of these requirements. These are faecal coliforms and faecal streptococci. Recently, workers have investigated Salmonellae and Ps. aeruginosa as possible indicator organisms.

Kenner and Clarke (1974) filtered large volumes of clear water and isolated, simultaneously, both Pseudomonas and Salmonellae. Over 75% of colonies with black centres from XLD agar proved to be

Salmonella Sp. Salmonella Sp recovery by other methods was very low due to the low survival of the organism in water. Pseudomonas was isolated from all samples but three. In this study, the authors observed that chances of isolating Salmonella or Pseudomonas increased as the quality of water increased.

Cherry et al. (1972) investigated the use if Salmonella Sp as indicators of water pollution, by a polyvalent fluorescent antibody reagent method. They obtained 60% more positive specimens than by using cultural methods. From moderately polluted waters, 65% of specimens were positive and 38% of specimens were positive in polluted water. They also found that 44% of the samples from streams, though to be unpolluted, were positive for Salmonella Sp.

Chau and Huang (1976) found that most enteric bacteria, apart from Salmonella, showed retarded motility in a semi-solid modium containing triphenyl methane, divalent cations and a low pH. They, therefore, developed a semi-solid enriched medium which they successfully used to screen for Salmonella. The slide agglutination test was used to confirm the presence of Salmonella.

Harvey and Price (1976) worked on sewage polluted river water and isolated Salmonella with Selenite F broth and a modified Muller-Kauffman tetrathionate broth. They found Selenite F broth more efficient in isolating salmonella. They inoculated the media directly with samples using equal volumes of broth and water.

The use of Ps.aeruginosa has been largely limited to recreational water quality analysis, although certain workers have tried to adapt it to drinking water analysis.

Hoadley (1968) wrote on the significance of Ps.aeruginosa in surface waters. He noted that the principal source of the organism is sewage and that surface waters may receive a considerable inoculum from storm drainage in municipal areas. The significance of the presence of Ps. aeruginosa in water may reflect the influence of drainage from inhabited areas, and of sewage discharges.

Carson and co-workers (1975) investigated factors influencing detection and enumeration of Pseudomonas by MPN and MF techniques. The type of filter used as well as the pH of assay media greatly influenced both recovery and development of characteristic colonial morphologies of the organism. The nature of diluents also exerted an influence.

Some authors have compared both Ps. aeruginosa and Salmonellae, with E. coli in a bid to show the value of the pathogens as pollution indicators. Two such works are reviewed below. Reitler and Seligmann (1957) discovered that both E. coli and Ps.aeruginosa can co-exist in pure water but that Pseudomonas exerted an inhibition on E. coli in late stage of growth, when placed together in peptone water. There was a critical range of peptone concentration at which this occured. In heavily polluted waters, this type of inhibition may occur possibly at time of sampling. The authors then suggested that Pseudomonas

should be considered along with coliforms in water quality analysis. This is because of the faecal origin and possible pathogenicity of Ps.seruginosa.

Van Donsel and Geldreich (1971) examined the relationship between Salmonellae and faecal coliforms in bottom sediments. Citing Dunlop et al. (1952), they stated that there was a ratio of one salmonella to 255,000 coliforms and to 4,800 enterococci in irrigation waters. Van Donsel and Geldreich (1971) detected one Salmonella organism in mud to every 150 coliforms in overlying water. The authors however expressed doubts as to the validity of their findings because in salmonella isolation from mud, some limitations on recovery rates are exerted. For example, certain types of bottom materials, especially clay, adversely affected the efficiency of the enrichment broth. This effect is presumably due to removal of essential ions or nutrients by clay.

Use of Faecal Coliforms As Indicators Of Pollution:

The coliform group comprising E. coli, Enterobacter, Klebsiella etc., has been used for a long time as an index of water pollution. Perhaps, the most outstanding properties of this group are their survival rate in water, and their logarithmic numbers in faeces. These organisms satisfy most if not all, of the requirements of a good indicator organism. However, the presence of coliforms in water may result either from faecal origin or from other sources. Therefore, the faecal coliform test is a more significant and reliable test than

the total coliform test since it indicates that pollution is directly or indirectly of faecal origin. There is abundant literature on the use of coliforms as an index of water pollution. Only a selected number will reviewed here. Many of the recent analysis are done by membrane filteration coupled with growth on a variety of media.

Kabler, (1954) worked on the purification and sanitary control variety of both potable and waste water. In his presentation on coliform organisms, he stressed that in the interpretation of coliform densities or classifications by types and species, careful consideration should be given to the methods used by investigators. He used a MPN procedure as against the multiple tube method commonly used before 1951 by investigators, On faecal coliforms, he asserted that E. coli type 1 (IMVIC ++ --) is of intestinal origin and so an indicator of faecal pollution. Other coliforms, he says, have been associated with vegetation,and soil. Therefore, the presence of these coliforms in water can have only questionable sanitary significance.

Grimes, (1975) investigated the release of sediment-bound faecal coliforms by dredging. The work was done during summer in the vicinity of Crosby Island where dredging operations were being carried out. Samples were taken before dredging, and after dredging. They found that faecal coliform concentrations increased significantly in the immediate vicinity of dredging operation. Such increased counts were attributed to the disturbances and relocation of bottom sediments by dredging, and a concomitant release of coliforms bound to bottom

sediments. They concluded that the observed faecal coliform densities probably did not indicate a health hazard. Highest counts were 200 per 100 ml of water. The implication, however, was that dredging of bottom sediments heavily contaminated with enteric pathogens could produce temporary health hazard down stream.

/ Deaner and Kerri (1969) carried out studies on the regrowth of faecal coliforms in sewage effluents. Quoting Geldreich (1967) in "Faecal coliform concepts in stream pollution", they reasonsed that only the non-faecal coliform portion of total coliforms could survive and multiply in water containing minimal concentrations of nutrients. The faecal coliforms require more favourable conditions, such as water temperatures above 15°C, bacterial nutrient source in concentrations above critical levels and adequate flow time between entry of discharge and the studied location downstream.

/ In their work, Deaner and Kerri (1969) did not observe any regrowth of faecal coliforms from sampling on an American river located near Sacramento and receiving effluents from a treatment plant. The fresh water is used for recreation, irrigation and as a source of drinking water. The sewage however was treated and there was short travel time between the outfall and the sampling stations. The river was shallow, swift and had a low turbidity.

There are indications of the existence of antibiotic resistant strains of coliforms. The danger posed by such resistant strains can not be overestimated. Cooke (1975), isolated coliforms and faecal

Coliforms from mussels, sewage, seawater and marine shell-fish. In 1976, she isolated these organisms from natural waters and other effluents. She studied their antibiotic resistance patterns. Mussels obtained from Rotoa, Protoiti and Brunner Lakes in South Island, N. Zealand contained coliforms and faecal coliforms resistant to one or more antibiotics. More than 70% of coliforms and faecal coliforms, collected over a 2-year period around a marine sewage outfull, were resistant to one or more antibiotics. 45% of those resistant to tetracycline or streptomycin could transfer this property to a susceptible E. coli K - 12. When coliforms and faecal coliforms were isolated from potable water supplies, as wells, springs, streams, seawater and effluents, and were examined for antibiotic resistance, she found more than 40% of the organisms from drinking water resistant to one or more antibiotics. In streams and seawater isolates, resistance increased to more than 60%. In effluents it was more than 70%. Up to 40% of all those resistant to streptomycin, or tetracyclin could transfer their resistance determinants to a susceptible E. coli K - 12 within 2-hours. Hence, although the widespread use of antibiotics may be responsible for increasing incidence of antibiotic resistant bacteria, these resistant faecal coliforms may have a selective advantage in natural waters, effluents etc.

Chlorination of effluents is practised in many countries in an effort to remore potential hazards of bacteria released with effluents.

The effect of chlorination on coliforms and faecal coliforms has been examined by several investigators. Silvey et al. (1974) studied the bacteriology of chlorinated and unchlorinated effluents in an 18-month period. In their results, they observed that treated wastewater effluents contained the greatest numbers of total and faecal coliforms. Chlorination was effective in reducing the numbers of micro-organisms only at stations where chlorine was directly applied. Bacterial populations recovered, as soon as chlorination was discontinued. Beneficial heterotrophic organisms were adversely affected by chlorination while faecal and non-faecal coliforms and faecal streptococci were less susceptible to chlorine. Chlorination did not totally destroy Salmonellae which were isolated with same frequency during periods of chlorination and non-chlorination. From their data, chlorination did not improve the pre-existing conditions of the river under study.

Davis and Keen (1974), examined municipal wastewater bacteria capable of surviving chlorination. Using selected diagnostic biochemical media for identification of bacteria they determined that a majority of species which re-established their populations within 21 days following chlorination, were lactose non-fermenters, not included in the Enterobacteriaceae. Many of these could be pathogenic and constitute a threat to public health in receiving waters. The authors observed that populations of coliform bacteria which survive chlorination apparently become attenuated and remain in that condition until the removal of the inactivating power of chlorine, from the environment.

Given enough time, adequate nutrients and other favourable conditions of growth, some coliform bacteria will regenerate their populations which may exceed their original order of magnitude.

Milbauer and Grosswicz (1959) worked on reactivation of chlorine inactivated E. coli. Using a concentrated solution of 10,000 to 15,000ppm of Naocl, they exposed suspensions of cells in either minimal medium or phosphate buffer for selected time intervals and in various media. They found that reactivation of chlorine treated cells was possible if cells are plated immediately after treatment on rich media. The method used for disinfection was found to affect the reactivation of cells.

Shuval et al. (1972) observed chlorinated effluents both in the field and in the laboratory. In their experiments, faecal coliforms were capable of growing in chlorinated effluents. When the effluent was stored for 3 days in a reservoir, the rate of regrowth correlated inversely to the residual chlorine in the reservoir, and to the number of coliforms surviving chlorination.

In the laboratory experiments, there were regrowths after treatment of effluent water with 11ppm chlorine. The regrowth occured in the laboratory even without inactivating the chlorine with sodium thiosulphate. These results suggest that the value of coliforms as an indicator of chlorinated effluents' pollution is difficult to assess. Both coliforms and faecal coliforms are capable of growing after chlorination of sewage effluents.

Brasswell and Hoadley, (1974), considered the recovery of E. coli from chlorinated secondary sewage. Using E. coli ATCC 27622 which was injured during chlorination of secondary sewage, they found the organism incapable of forming colonies on membrane filters incubated on M.Fc medium. The cells neither grew nor produced gas in lactose both. In concluding, they suggested that standard methods for examining effluents for faecal coliforms should not be applied to chlorinated effluents, since they could give false negative results.

A number of bacteriologists have worked on the possibility of purifying effluents by filteration through soils, before being discharged into water bodies. Krone et al. (1955) and Pound and Crites (1973) reported that when wastewaters are passed through a meter of soil, most bacteria are removed. Many do not penetrate beyond a few centimeters of soil. The removal was due to mechanical straining and sedimentation on the surfaces of soil particles.

Mallmann and Litsky (1951) found that coliform bacteria survived longer in loam and muck soils than in sandy soils. Van Donsel et al. (1967) also found, that faecal coliform bacteria (E.coli) survival varied seasonally.

To this end, Edmonds, . (1976) set out to determine the environmental soundness of utilising dehydrated sludge on clearcut forest areas and on graveled glacial outwash soils. He sought to determine the length of survival time of total and faecal coliforms in the sludge and soil systems, in relation to environmental conditions,

and to determine whether faecal coliforms in sludge are capable of moving into the soil, into groundwater and eventually into streams. Total and faecal bacteria moved from sludge into soil but only a few penetrated beyong 5cm. The soil is an effective bio-filter as only few faecal bacteria occured in the groundwater. Counts were between 5 and 52/100 ml. Groundwater contamination from vertically moving pathogens was unlikely though there is a hazard from surface run-offs.

Methods And Media Used For Counting Coliforms:

There are several methods and media available for the enumeration of coliforms and faecal coliforms in the environment. In many cases, media are tied to specific methods. Consequently, there is often a problem of choosing what combination of media and methods to be used in isolating and enumerating faecal coliforms in water analysis. A few media and methods are reviewed in this presentation.

Thomas and Woodward (1955) estimated coliform densities by the MF and fermentation tube procedures. The MF technique was 2-5 times more precise than the MPN procedure, which gave higher estimates of coliform densities. The observed disparity was attributed to the mathematical nature of the MPN method which often tends to overestimate the true coliform density.

Allen et al. (1952) worked on conditions affecting growth of E. coli on bile salt media and its enumeration in polluted waters. They used MacConkey's agar containing peptone, sodium taurocholate, lactose and 1% neutral red; a medium similar to this but containing

5g/ of specially pure bile salts; another containing 20g/ of ox-gall instead of sodium taurocholate; and Hajna and Perry's EC-medium containing neutral red as indicator. The presence of bile salts and lactose in the medium inhibited growth at 37°C, This inhibitory effect increased for most strains at 44°C. With the addition of inorganic salts as phosphates to the media, inhibition increased and some strains failed to grow. This formed the basis for developing a technique for counting bacteria, adaptable to polluted waters.

Dukta and Tobin (1976) evaluated the efficiency of four procedures for enumerating coliforms in water. These procedures were the MPN technique using lauryltryptose broth (LST) and 2% BGB; MF procedure using M-endo agar LES; MPN with FCDS and BGB developed for Indian freshwaters; and an MF technique using MC-agar developed for seawater. Their data indicate that within the sampling area, maximum population estimates were achieved with the MF procedure using M-Endo agar. Each of the methods was selective for isolating different genera. The authors concluded that the possibility of a universal coliform estimation test is remote and that local conditions would dictate the success of a particular procedure for isolating and counting coliforms.

Pugsley et al. (1973) proposed a new medium for detecting gas and indole in a single tube. They compared this medium with a method used in the 44°C E. coli confirmatory test. The inclusion of mannitol in their medium reduced the number of false gas reactions. Addition of tryptone in the medium made it possible to test for both indole

production and mannito- fermentation in the same tube. Results were obtainable in 24 hours and the method was fast, easy to use and inexpensive.

Geldreich et al. (1965) employed M-Fc medium and the MF technique in isolating and counting faecal coliforms. They compared results obtained by this method with those of the MPN confirmatory test for faecal coliforms with 44.5°C inucubation. The results obtained fell within the 95% confidence limit. While lactose fermentation in the MF technique is measured by the production of acids, gas production is used as evidence of fermentation in the MPN technique. 93.2% of blue colonies growing on M - FC medium were faecal coliforms. The authors believed that the two techniques were measuring the same organisms.

Standard Methods for the Examination of Water and Wastwater (APHA 14th edition 1976) recommends the use of endo-type media and the MF technique in detecting and counting coliforms. Any colony growing on the medium with any amount of metallic sheen is to be regarded as a coliform. Eosin - methylene blue agar may be adapted for use in the direct count method of Clarke, (1971). This method remains the most accurate means of counting bacteria. In water samples, accuracy of counts by the MF technique depends on the level of pollution in the water. Only low to mederately polluted waters can be analysed by the method with reasonable accuracy. The MPN procedure is statistical in nature and so, gives only an estimate of the true bacterial density.

The direct count method with EMB seemed appropriate for this research since the samples are expected to be heavily polluted, and because true bacterial numbers and not estimates are desired.

Use of Faecal Streptococci As An Indicator Of Water Pollution:

A progress report was compiled by the Committee on public health activities, (committee on public health activities 1961). The view was advanced that coliform density determinations are not fully acceptable, because the sanitary quality of waters is dependent on the numbers of faecal E. coli present and not on numbers of total coliforms. The coliform test, although inadequate, is still frequently used. Attempts have been made to use a variety of bacteria as pollution indicators, but none of these have been upheld as a sole indicator of water pollution. The report mentioned the work of Heukelekian and Rosenberg, who have compared coliforms and enterococci as indicators of well water pollution. Based on their results, they were uncertain as to whether E. coli should continue to be used as water quality index since recent advances in media development have made it easier to perform enterococci analysis. However, other workers are hesitant in accepting faecal streptococci as the sole indicator of pollution. The report stated that there is good evidence that faecal streptococci survival rate is less than that of coliforms in unpolluted water. Experiments on faecal streptococci viability in water show that the organisms die out more rapidly than do coliforms. Gilcreas and Kelly were quoted in the report as saying that the coliformstest was still the most reliable test in determining any significant pollution.

The direct count method with EMB seemed appropriate for this research since the samples are expected to be heavily polluted, and because true bacterial numbers and not estimates are desired.

Use Of Faecal Streptococci As An Indicator Of Water Pollution;

A progress report was compiled by the Committee on public health activities. (committee on public health activities 1961). The view was advanced that coliform density determinations are not fully acceptable, because the sanitary quality of waters is dependent on the numbers of faecal E. coli present and not on numbers of total coliforms. The coliform test, although inadequate, is still frequently used. Attempts have been made to use a variety of bacteria as pollution indicators, but none of these have been upheld as a sole indicator of water pollution. The report mentioned the work of Heukelekian and Rosenberg, who have compared coliforms and enterococci as indicators of well water pollution. Based on their results, they were uncertain as to whether E.coli should continue to be used as water quality index since recent advances in media development have made it easier to perform enterococci analysis. However, other workers are hesitant in accepting faecal streptococci as the sole indicator of pollution. The report stated that there is good evidence that faecal streptococci survival rate is less than that of coliforms in unpolluted water. Experiments on faecal streptococci viability in water show that the organisms die out more rapidly than do coliforms. Gilcreas and Kelly were quoted in the report as saying that the coliform test was still the most reliable test in determining any significant pollution.

The committee members considered the coliform test satisfactory for measuring sanitary quality of municipal water supplies.

The view that faecal streptococci should replace coliforms as the index of water pollution is upheld by many bacteriologists. Workers like Abshire and Guthrie (1971), Geldreich and Kenner (1969), Morris and Weaver (1954), Sayler et al. (1975) and Allen et al. (1953), support the use of faecal streptococci as pollution indicators. Some of these advocate that faecal coliforms and faecal streptococci should supplement each other in assessing water quality.

Poor Acceptance Of Faecal Streptococci Index:

The reason for the poor acceptance of S.faecalis as an indicator of water quality was examined by Kenner et al. (1961). The poor acceptance, they claimed, is due to the relatively low recovery rates of the organism in comparison to coliforms; multiplicity of detection methods and poor agreement between various quantitative enumeration methods; and lack of detailed systematic studies on the sources, survival and interpretation of S. faecalis in various types of waters. There are other sub-groups of the organism,as S. bovis and S. equinus which are not found in human faeces. These are considered as specific indicators of animal pollution. They die quickly outside the gut of animals and their isolation indicates recent contamination.

Geldreich et al. (1969) studied 12,536 strains of faecal streptococci from faeces of warm-blooded animals and many water sources in a

vegetation. Because of these findings, the need arose to include faecal coliforms in recreational water quality determinations. This procedure helped confirm the source of the faecal streptococci in densities below 100/100 ml of water.

Advantages of Faecal Streptococci As an Indicator.

Sayler et al. (1975) collected samples from five different stations in the upper Chesapeaks bay. The indicator bacteria were distributed in the water, independent of temperature and salinity. Counts were not correlated with the concentration of suspended solids. However, 53% of the total viable bacteria, and more than 80% of faecal indicator bacteria were directly associated with suspended sediments. They observed prolonged survival of faecal streptococci in sediment samples. Their results compare favourably with FC:FS ratios for various other samples.

Allen et al.(1952) investigated some factors responsible for viability of faecal bacteria in water. They showed that the mortality curves of both B. coli and S. faecalis were characteristic for each of the organisms. They were both less viable at pH 6-7. S.faecalis was not affected by the degree of oxygenation of the water but B.coli died much more rapidly under anaerobic than aerobic conditions. B.coli grew at organic matter content as low as 0.28ppm, while S.faecalis required much higher concentrations of organic matter to show any appreciable growth. This result of would suggest that streptococci would be a better indicator of drinking water quality.

Morris and Weaver (1954) sampled 54 wells from which duplicate

samples were collected. One of each pair was analysed within six hours and the other after 24 hours in dark storage. They used the MPN technique to determine both coliforms and streptococcus densities. There was close agreement in densities of the two organisms in the 6 hours samples. Streptococci did not grow in the stored samples and the coliform counts were erratic. There were increases in numbers in some samples and reductions in most. These observations tallied with those of Hanes et al. (1964) and Leininger and McClesky (1953).

Hanes et al (1964) investigated the influence of dissolved oxygen (D.O) on the survival of coliforms and streptococci. High D.O. levels stimulated a 5.7 times increase in numbers of coliforms. Normal D.O. levels brought about the smallest increase in numbers while low D.O. caused a 5.1 times increase in cell numbers. With enterococci, only at low D.O. did a small and apparent increase in numbers occur. The increase was so small that it was discounted as growth. Because the death rate of enterococci is greater than that of coliforms, the authors concluded, that on the basis of their results, enterococci seem to be better indicators of recent faecal contamination of water than coliforms.

Use of Selective Media and Diluents.

Bissennette et al. (1975) enumerated indicator bacteria from natural waters. They observed that exposure to the aquatic environment caused a significant proportion of cells to lose colony-producing ability on selective media. This did not occur in nutritionally rich non-selective media. Discrepancies in colony-forming units between non-selective and selective media showed that a substantial portion of cells may have become injured. The injury could have been due to the stress imposed by the aquatic environment. The extent of injury varied from water to water.

Hoadley and Cheng (1973) selectively recovered indicator bacteria. They used trypticase-soy agar and other selective media. Tap water was found to be highly toxic to all strains of Pseudomonas, E coli and S. faecalis. Phosphate buffer was a more successful diluent for Ps. aeruginosa. The buffer solution did not improve on E. coli recovery and S. faecalis remained viable after suspension in double distilled water or phosphate buffer. All organisms grew in 0.1% peptone water or stream water following a lag period of varying lengths of time. When suspended in aqueous media, injury occured which prevented recovery of viable cells on selective media including those that would normally grow the organisms. There is a need to reduce selectivity of media used in water analysis.

On selection of diluent for bacterialogical examinations, Butterfield (1932) observed that phosphate water and formula C water gave the most consistent results. Diluents for analysing natural waters, he claims, must contain some mineral salts. pH should not be lower than 8.2. At this pH survival of

after 5 to 32 months at different temperatures. In distilled water, viability was retained for periods of 14 to 32 months. B. typhosa survived better in distilled water than in 0.85% NaCl. The same was true for B. coli, B. tuberculosis, B. diphtheriae, S. haemoliticus and S. viridans. Room temperature appeared more suitable for the survival of most bacteria in distilled water. Streptococci and B. diphtheriae are exceptions.

Media And Methods For Streptococci Analysis

Kabler, (1954) compared the MPN and MF procedures on the basis of 95% confidence limits of the confirmed MPN. There was a 73.8% agreement of 1706 samples collected and examined simultaneously. Of 650 samples, there were 114 ± agreement in results 457 negatives agreed and 79 disagreed. Results of 88% of samples analysed by the two methods agreed. Apart from the turbidity of samples, there was no other readily apparent relationship between agreement of results by the MPN and MF methods and any recorded physicochemical characteristics of samples. These results indicate that the two test procedures do not measure the same group of organisms. The sanitary significance of this anomaly is yet to be determined.

Pugsley and Evison, (1974) developed the technique of Abshire and Guthrie (1971), and of Pavlova et al. (1972). They combined the FA test with MF technique. They designated the new procedure the MF - FA test. Positively stained micro-colonies were clearly visible on filter surfaces as brightly fluorescent areas. They eliminated the possibility of false

negatives by conducting control experiments with other bacteria than group D streptococci, using selective incubation medium and trypsinisation of micro-colonies.

On methodology, Abshire and Guthrie (1971) used a slide method with fluerescent antibody technique, for detecting S. faecalis. The method permitted S. faecalis of faecal origin to be distinguished from those of soil etc. Their reactions were specific and confirmed by biochemical tests. The results showed that the method is useful in pollution studies requiring rapid identification of polluting organisms.

Daoust and Litsky (1975) enumerated faecal streptococci by the MF technique using Pfizer selective enterococcus (PSE) agar overlay method. They also used M-enterococcus medium, and KF streptococci medium. Isolation of streptococci with KF medium and M-enterococcus agar were similar with faecal samples. PSE showed highest counts with both faecal and marine water samples.

Brodsky and Schiemann (1975) used PSE agar and KF medium in counting streptococci from water, by the MF technique. PSE was highly selective for the enterococci. The tan colour resulting from esculin hydrolysis was found unnecessary because over 90% colonies isolated on PSE agar were confirmed as faecal streptococci. KF agar recovered significantly greater numbers of colonies but selectivity was limited to 83% typical streptococci, and 54% enterococci. The 45°C incubation of KF agar to improve its selectivity was unsuccessful.

Wie-shing Lee (1972), using PSE agar for screening, devised tyrosine

decarboxylase (TD) medium and D-broth for additional confirmation and identification of group D enterococci. By this method, 97% of enterococci could be accurately identified and confirmed.

Cooper and Ramadan (1955) using heat, tetrathionate, tellurite and thallium salts, differentiated between human and animal faecal streptococcal pollution. The tellurite salt method proved highly efficient in isolating 97% of strains from human, cow and sheep faeces. Heat resistance differential tests divided isolates into groups some of which were source specific. This method may be useful in tracing water pollution to its source.

Allen et al. (1953) enumerated streptococci in polluted waters. Their apparatus was similar to those Allen et al. (1952), but included glucose-yeast extract-sodium azide agar. The 45°C incubation temperature they employed, coupled with the presence of azide reduced the number of colony-forming cells. Attenuated cultures were more affected by this inhibition. The incorporation of neutral red in the media was so inhibitory that it had to be excluded.

Faecal streptococci were not used until 1971 in the U.S.A. as an index of water pollution. Its use was however practised in Great Britain before this time - (Abshire and Guthrie, 1971). Slanetz and Bartley (1964) called for its use in the U.S.A. The evidence they gathered suggested that the faecal streptococci are more practical and reliable indicators of faecal contamination of water than faecal coliforms. The authors highlighted that M-enterococcus agar is 100% selective for faecal streptococci and that this procedure had been

adopted at the Pasteur Institute in Madagascar. For routine water analysis, that Standard Methods (11th edition 1960) included a multiple tube dilution method using AD-EVA broth, and a MF technique using M-enterococcus agar, for faecal streptococci detection. They found that the KF-medium was not as selective as M-enterococcus agar was for faecal streptococci. KF-medium gave poor growths of S. bovis. Contamination due to pseudomonads, gram-negative rods and micrococci found on KF-medium was absent with M-enterococcus agar. The authors drew attention to Cooper and Ramadan (1955); Bartley and Slanetz (1960); and to Kenner et al. (1960) who all pointed out the value of the faecal streptococci index in tracing source of water pollution. They agree, however, that there is need for more studies on the method. The presence of faecal streptococci in water is positive evidence of faecal contamination; and relatively simple detection and enumeration methods are available, they argued.

Slanetz and Bartley (1957) formulated the medium M-enterococcus agar which they used to dtermine number of enterococci by the MF technique. The medium yielded better results by this method than other available media. Colonies were larger and counts higher. Selectivity was 100%.

Clark , (1971) studied plate preparations, inoculum volume and technique of spreading, in the surface plate method of counting bacteria. He showed that predrying plates to a weight loss of 3 - 4g/15ml allowed the use of inoculum volumes of up to 1 ml, without affecting bacteria

recovery. Plates were stacked or separately placed, upside down at 24° - 50°C for drying. Spreading the inoculum to its complete absorption by the agar or only for 15 seconds gave equally good recoveries of the bacteria.

The term "coliforms" as used in this presentation is defined as applied to MF technique found in Standard Methods, (APHA 1976) which comprises all aerobic and facultatively a aerobic gram-negative, non-spore-forming, rod-shaped bacteria that produce a dark colony with metallic sheen within 24 hours on an endo-type medium containing lactose.

The term 'faecal coliforms" is used for coliforms able to grow and produce gas in brilliat green bile lactose (BGBL) broth at 37°C, and produce gas in lactose broth at 44.5°C.

The term "faecal streptococci" includes gram-positive cocci appearing in chains of four or more cells, that are able to grow on M-enterococcus agar, forming pink, red or maroon colonies; and which, when placed in ethyl-violet azide (EVA) broth, will form a purple button growth in 48 hours at 37°C. The direct count method was preferred in this research because of its accuracy and simplicity. Pollution in effluents was high and the method offered a better means of counting the high numbers of bactoria, than would the MF technique. The MPN method was not used because of its inherrent mathematical problem. The procedure of Clark (1971) was followed closely during drying of plates and inoculation. Isolation and counting of faecal coliforms was carried out on EMB in conformity with Standard Methods (1976)

MATERIALS AND METHOD

Media.

M-enterococcus agar (M-EA), Eosin-methylene blue (EMB) agar Triple-Sugar iron (TSI) agar Ethyl-violet azide (EVA) broth; Brilliant green bile lactose (BGBL) broth Selenite F broth and Salmonella-Shigella (S-S) agar were used. M-EA, EVA and Selenite F were manufactured by BBL. TSI, EMB, BGBL, and SS media were manufactured by Oxoid.

GLASSWARE:

Sterile 10 ml pipettes; sterile 1 ml pipettes, sterile cotton-plugged test-tubes; sterile wide - mouth stoppered bottles and sterile bent glass rods were used.

PREPARATION OF MEDIA:

All media were prepared from dehydrated powders according to manufacturer's instructions. They were all incubated for 24 hours at 37°C and sterility checked prior to use. Only freshly prepared media were used throughout the examination.

SAMPLING SITES:

Samples were collected from various sites in the environs of the Kubanni Lake, Kubanni River and sewage treatment plant at A.B.U. Main Campus, Zaria. (Fig.1).

The following sites were examined:

Site A, point of entry of Samaru stream into Kubanni Lake;

Site B, intake structure to the proposed water purification works

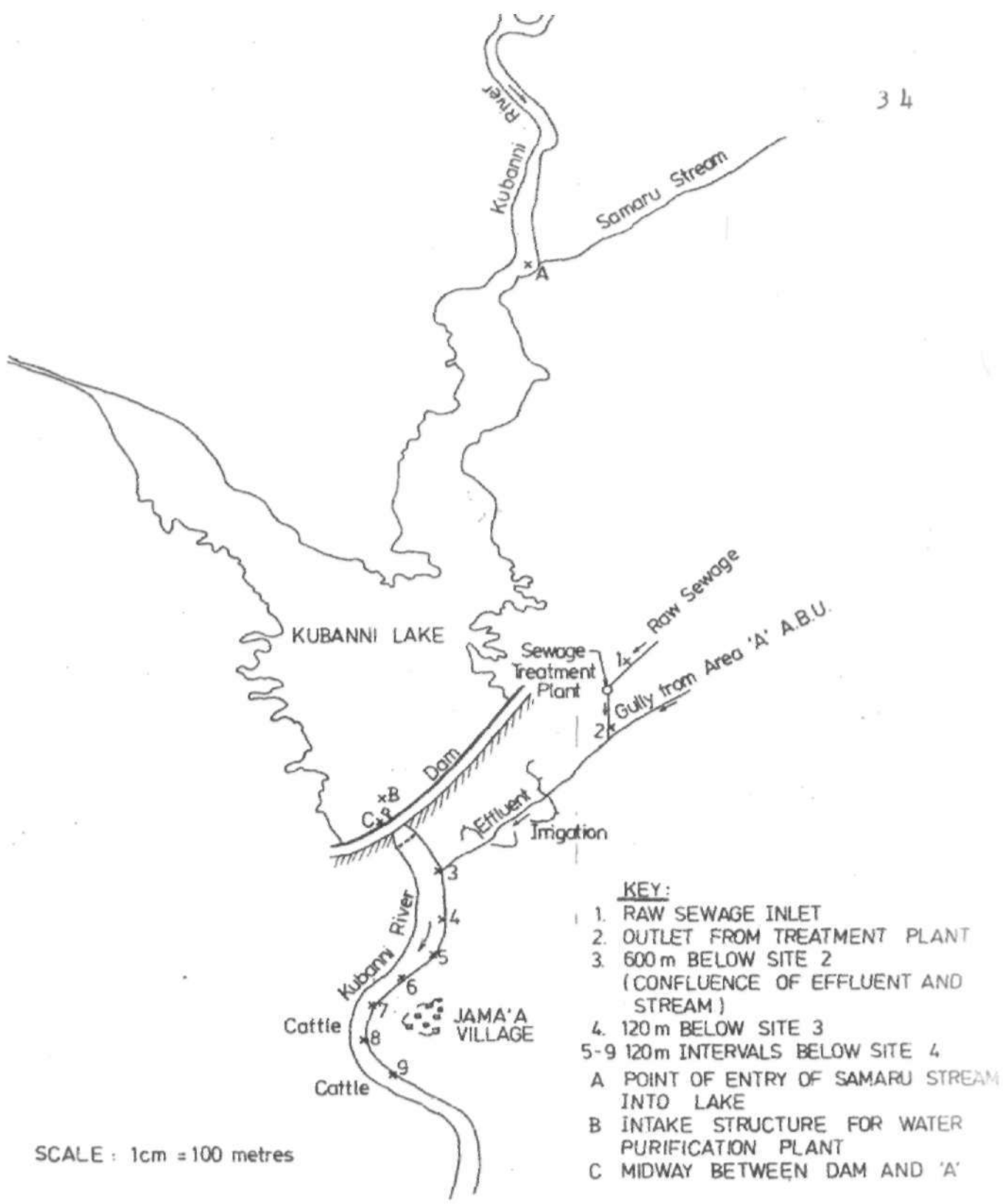


FIG.1 GENERAL LOCATIONS MAP OF SAMPLING STATIONS.

for A.B.U. main campus; Site C midway between the dam wall and spillway Sampling sites along effluent gully and Kubanni River were site 1, raw sewage influent to the sewage treatment plant; Site 2, sewage effluent outlet from the treatment plant; Site 3, confluence of sewage plant effluent and Kubanni River; and site 4, about 100 meters downstream from site 3. Additional sampling sites (5-9) at 120 meter intervals downstream of site 4 on the Kubanni River were added later. Samples were taken for the period April - January 1978 to represent both raining and dry season adequately.

COLLECTION OF SAMPLES:

All samples were collected in sterile, wide-mouth, stoppered 500 ml bottles between 9.30 am and 10.30 a.m. on each sampling day. This period represents period of peak flow of sewage at A.B.U. Samples were collected once a week. Samples from sites B and C were collected by lowering the bottles into the water with the aid of a piece of twine tied around the neck of the bottles from a platform. All other samples were obtained by dipping the bottles under water with their mouths facing the direction of flow. The top third of the water in the bottles was always discarded and the bottles stoppered immediately. The outside of all sampling bottles were disinfected with concentrated 'Dettol' solution immediately after each collection. The portions of twine coming in contact with water or sewage were out-off and discarded. Rubber gloves used for sample collection were also subjected to disinfection with 'Dettol' after each collection. Samples were not packed in ice during transportation to the laboratory for analysis because distances between the sites and the

ANALYSIS OF SAMPLES:

The samples were analysed soon after collection. In one or two cases when such was not possible, samples were stored in a refrigerator but not for more than one hour.

Serial Dilution:

Ten-fold dilutions were made from each sample after mixing by shaking 25 times (A.P.H.A. Standard Methods, 1971). Dilutions ranged from 10^{0}-10^{6}. The tubes were appropriately labelled and serially arranged on a test-tube rack. The tubes of serial dilutions were cotton-wool plugged as were those containing sterile distilled water. Contamination of the samples was prevented by maintaining aseptic conditions throughout sample analysis.

Plating:

As soon as the serial dilution of any sample was completed, four consecutive dilutions were selected for plating. The criterion for selection was based on the expected level of pollution at the various sites. This was roughly determined by initial trial plating of samples at beginning of sample collection. After shaking the test-tubes, 0.1 ml was removed and transferred to the surface of EMB and M-EA in petri-dishes, using a fresh 1 ml pipette for each dilution. Duplicate plates were inoculated for every dilution.

The water was evenly spread on the agar plates using sterile L-shaped glass rods. Spreading was continued until most of the water was absorbed by the medium (Clark 1971). A fresh sterile glass rod was used for each plate and all plates were adequately labelled.

Incubation:

All plates were incubated under humid conditions at 37°C for 24 hours. M-EA plates were left in the incubator for 48 hours. After incubating for these times, plates were removed, colonies counted and their numbers per dilution recorded.

Reading of Plates:

The EMB plates were used to differentiate between lactose positive (pink ; lactose negative (white or colourless); and E. coli (metallic sheen) colonies. Each of these colony types was counted for every dilution and plate.

M-enterococcus agar was used to isolate faecal streptococci. All maroon, pink or red colonies found growing on the agar surface were counted as presumptive faecal streptococci (Slanetz and Bartley 1957).

Confirmatory Tests:

All coliforms and streptococci were subjected to confirmatory tests. All lactose positive, and colonies growing on EMB with a metallic sheen were confirmed by inoculating them into BGBL and lactose broths with durham tubes.

BGBL tubes were incubated at 35°C for 24 hours in a water bath. Tubes were read as positive if they had turbidity and gas. Negative tubes were incubated for a further 24 hours. Lactose broth was incubated at 44.5°C for 24 hours. Turbid tubes with gas were taken to be faecal coliforms according to Buttiaux et al. (1956); and Papavassiliou (1958). Loopfuls from positive tubes were streaked on EMB agar from both BGBL and lactose broths. Reproduction of pink or metallic sheen colonies was regarded as confirmatory for faecal coliforms.

All maroon, pink or colonies on M-EA were inoculated into EVA-broth in tubes. The tubes were incubated at 37°C for 48 hours. Development of a purple button growth after 48 hours was regarded as confirmatory for faecal streptococci (Sayler, et al. 1975). All positive tubes were gram-stained and observation of gram-positive cocci in chains of our or more cells was used as augmentation for the confirmatory test described above.

ISOLATION OF PATHOGENS:

Attempts were made to isolate a few pathogens (see fig.2). At the same time as samples were being analysed for faecal coliforms and faecal streptococci, 3 ml of undiluted sample from each site was inoculated into an equal volume of double strength selenite F broth in bijou bottles. All bottles were labelled and incubated for 24 hours at 37°C (Harvey and Price 1976). From these bottles, loopfuls

were streaked on S S agar in petri-dishes. The labelled agar plates were incubated at 37°C for 24 hours. Black centred colonies were randomly picked from the S S agar plates and transferred to TSI agar slants and incubated for 24 hours at 37°C. Presumptive Salmonella, Shigella, Proteus etc. identified on basis of TSI reaction, were subjected to biochemical tests using KCN broth, Simmon's citrate, urea agar, tryptone broth, lysin decarboxylase broth and oxidase test.

Salmonella, Shigella, Pseudomonas, Proteus and other enteric gram-negatives were identified in accordance with the method of Edwards and Ewing (1972). A scheme of analysis is presented in figure 2.

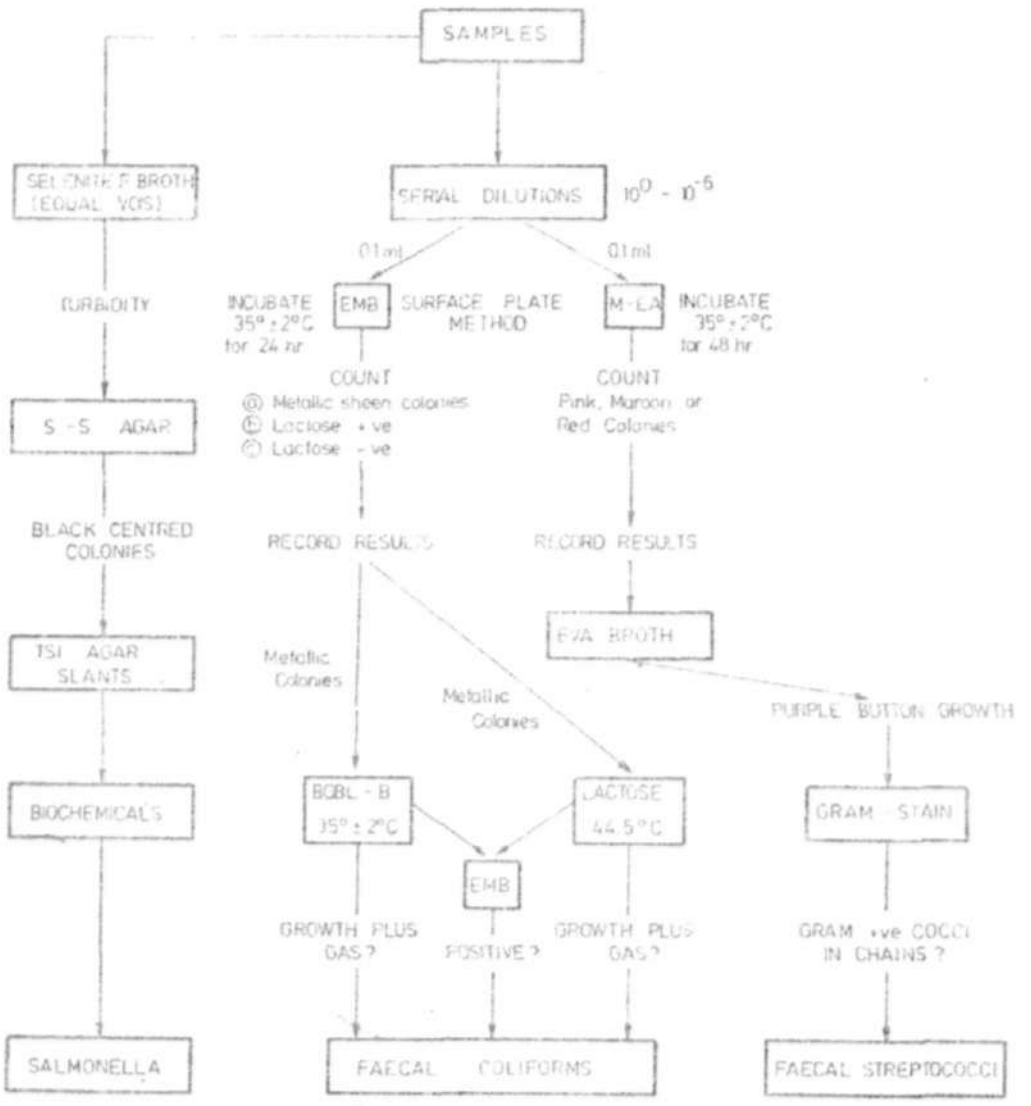


FIG 2 SCHEME OF ANALYSIS

RESULTS

Faecal Coliform Densities Over A 10-Month Period

Figure 3 is a plot of monthly mean faecal coliform densities at sites 1 - 4 for the 10-month sampling period (April, 1977 - January 1978). Counts were highest at site 1 for raw sewage (see fig. 1). The highest density occurred in April, 1977 and was 1 x 10^8/100ml while the lowest observed density was 2.5 x 10^6/100ml in August, 1977. After August, densities rose steadily and finally appeared to stabilize between September, 1977 and January, 1978.

Faecal coliform densities at site 2 showed only slight reductions in comparison to site 1 densities. In the rainy season months (April - August), counts at site 2 ranged from 1 x 10^6 per 100 ml to 3.5 x 10^6/100ml with a peak in July. There was a drop in August and a steady rise thereafter between September 1977 and January, 1978. The lowest monthly mean density of 1 x 10^6/100ml was in August while

FIG 3 FASCAL COLIFORM DENSITIES OVER A 3 MONTH PERIOD FOR SITES 1-4.

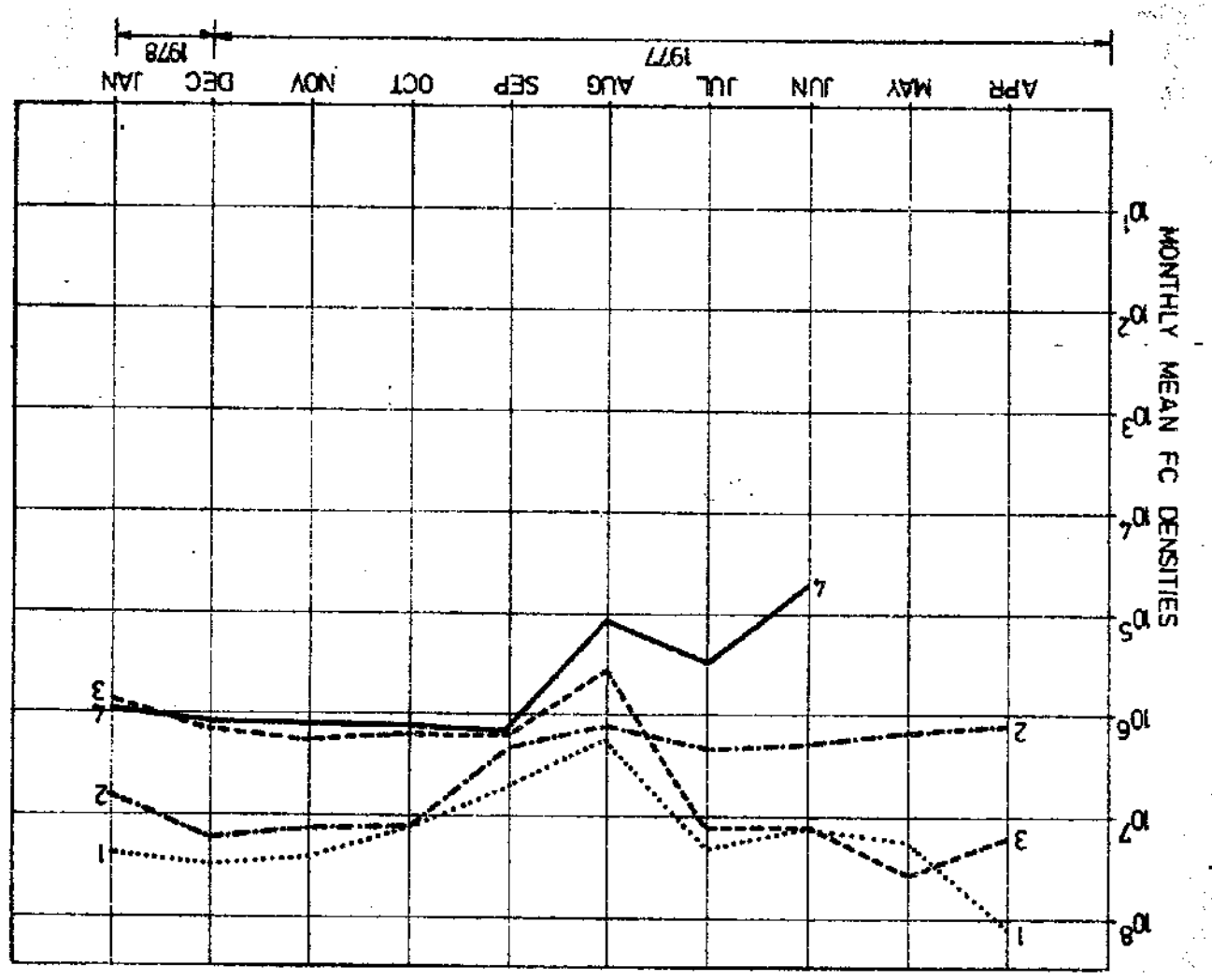


TABLE 1a: EFFICIENCY OF SEWAGE TREATMENT PLANT BETWEEN APRIL 1977 AND JANUARY 1978 IN REMOVING FAECAL COLIFORMS BETWEEN SITES 1 AND 2.

	1977 APRIL	MAY	JUNE	JULY	AUGUST	SEPT.	OCT.	NOV.	DEC.	1978 JAN.
SITE 1	$1x10^8$	$2.9x10^7$	$1.5x10^7$	$3.5x10^7$	$2.6x10^7$	$7.8x10^6$	$1x10^7$	$4.3x10^7$	$5x10^7$	$4x10^7$
SITE 2	$1x10^6$	$2.5x10^6$	$3x10^6$	$3.8x10^6$	$1x10^6$	$3.5x10^6$	$1.0x10^7$	$1.9x10^7$	$2.5x10^7$	$8.4x10^6$
% REDUCTION	1 99	91	80	89	96	55	0	56	50	79

the highest of 3 x 10^7/100ml was in December, 1977. From these results and figure 3, the treatment plant reduced faecal coliform numbers from 1 x 10^8/100ml in site 1, to 1 x 10^6/100ml in site 2 during April 1977. From April 1977 till January, 1978, reduction was small if at all. The reduction in faecal coliform numbers from site 1 to site 2 was minimal between August 1977 and January, 1978.

Site 3 showed rather high densities between April and July. There was again a drop in counts in August and a concomittant rise in September followed by a levelling off till January 1978. The curve is roughly parallel to those for sites 1 and 2 though densities were lower between July, 1977 and January, 1978. From April to July, densities at site 3 were high, even higher in some cases than was observed for sites 1 and 2. At site 4, faecal coliform densities were generally low during the rainy season. There was a drop in counts from July - August 1977, which rose again in September and levelled thereafter. Densities at site 4, appeared to be similar to site 3 densities from September, 1977 to January, 1978.

Although counts were lower at site 4, the differences from those at site 3 were very small. Even during the rainy season when dilution was at its highest due to overflow of the lake, reductions in coliform numbers was small. About 1,000 meters from the sewage disposal plant the lowest site 4 density during this period was still 2 x 10^4/100 ml. In the dry season, the reduction was less; the lowest site 4 count being 9.5 x 10^5/100ml in January.

Faecal Streptococcus Densities During Rainy And Dry Seasons:

In figure 4, faecal streptococci results for three sites (1 - 3), are compared for three consecutive rainy season and three consecutive dry season months. Faecal streptococci density reductions between sites 1 and 2 is minimal. Atsite 3, the reduction was still insignificant during the two seasons. Densities at sites 2 and 3 appeared to be higher during the dry season. The highest dry season count for site 1 was in November (2×10^7/100ml) while the highest observed rainy season density for the same site was 8.5×10^6/100ml in May.

At site 3, the lowest rainy season and lowest dry season counts were the same and it was 2×10^5/100ml for each (June 1977 and January, 1978). In the rainy season (May and July, 1977), the mean density for sites 2 and 3 were about the same. A similar observation was made in June and December, 1977 for sites 1 and 2.

Comparison Of Faecal Coliform Densities With Faecal Streptococcus Densities At One Site (4)

Faecal coliform and faecal streptococcus densities at site 4 were compared (Fig.5). Over a 16-week period during the dry season, faecal coliform and faecal streptococcus counts showed some degree of correlation. The faecal coliform densities were generally constant and higher whereas faecal streptococcus densities were not

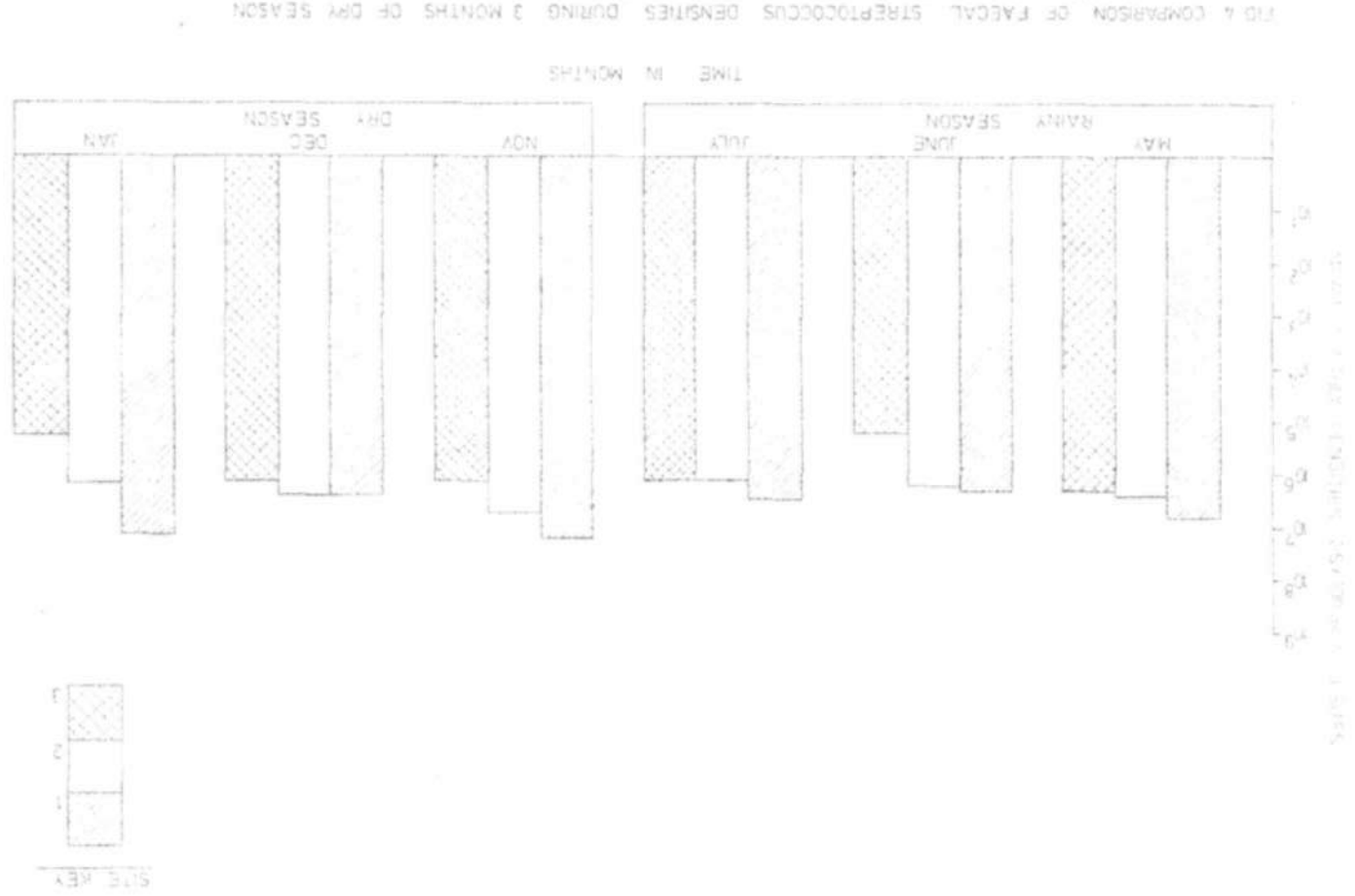


FIG 4 COMPARISON OF FAECAL STREPTOCOCCUS DENSITIES DURING 3 MONTHS OF DRY SEASON AND THREE MONTHS OF RAINY SEASON FOR SITES 1 - 3

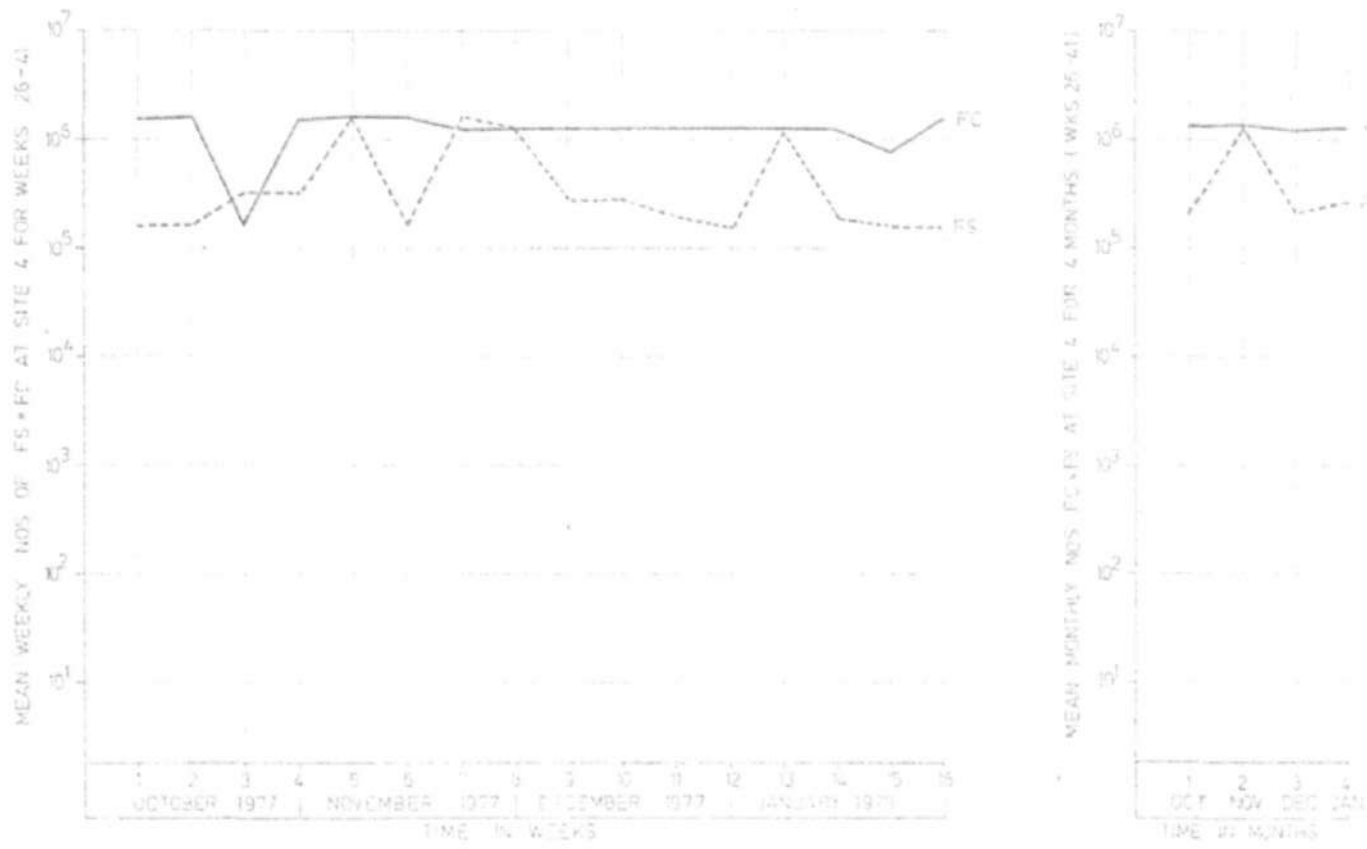


FIG 5 COMPARISON OF FAECAL COLIFORM (JUNGLES) WITH FAECAL STREPTOCOCCUS DENSITIES AT SITE 4 DURING 4 MONTHS OF DRY SEASON

uniform and lower. The fluctuations in faecal streptococcal counts occurred all through the 16-week period. Although faecal streptococcal numbers are lower than faecal coliform numbers, fig. 5 shows that their densities are comparable. Inset of fig.5 is the monthly means of the same organisms during the same period. Table 1 contains the mean 10-month densities for sites 1 - 4 for faecal coliforms and faecal streptococci. Faecal coliform and faecal streptococci densities appeared to be approximately the same from site to site. Between mid-April and last week of May, 1978, A.B.U. was closed and students were sent home following some disturbances. This was a unique opportunity to test coliform bacteria and streptococcus densities at all sites (1-9) when all students were off campus. Results of a set of samples taken during this period are presented in table 2. This shows the effect of the total absence of students from the campus on the treatment plant effluents. Densities at sites 1 and 2 were 3 x 10^5/100ml FC and 5 x 10^4/100ml FC respectively. Faecal coliform counts at sites 3 and 4 were both 1 x 10^3/100ml FC. Also, faecal coliforms continued to appear in samples until site 6 and disappeared thereafter at sites 7, 8 and 9. These last three sites are located in front of and on either sides of Jama'a village.

Faecal streptococcus counts for sites 1 and 2 during this period were 1 x 10^4/100ml and 1 x 10^4/100ml respectively. No faecal

TABLE 1b

Mean 10-months FC and FS Densities for Sites

1 - 4 And B and C

SITES	FAECAL COLIFORMS/100ml	FAECAL STREPT. /100ml
1	1.3×10^8	7.1×10^7
2	3.1×10^7	1.3×10^7
3	1.9×10^7	4.9×10^6
4	3.2×10^6	2.8×10^5
B	2.2×10^2	1×10^1
C	8.2×10^2	1.1×10^1

TABLE 2

Densities of faecal coliforms and faecal streptococci during absence of students from campus. From Mid-April-May Ending 1978.

Site	Faecal Coliforms/100ml	Faecal Streptococci/100ml
1	3×10^5	1×10^4
2	5×10^4	1×10^4
3	1×10^3	-
4	1×10^3	-
5	2×10^1	-
6	-	-
7	-	-
8	-	-
9	-	-

- = . Negative counts

streptococci were detected from sites 3 - 9 at this time. At no other occasion during the 10-month research period were negative cultures obtained from any of the 9 sites. Results were different for the same sites (5-9) during the dry season. The results of dry season (sites 5 - 9) samples are included in stable 3. After 600 meters downstream of sire 4, samples from site 9 still gave faecal coliform counts of 2.3 x 10^3/100ml and faecal streptococcus counts of 1 x 10^3/100 ml. This stretch passes in front of Jama'a village and extends about 220 meters beyond it.

SAMPLING OF THREE SITES ON THE KUBANNI LAKE

The potability of water in Kubanni Lake was examined through sampling sites A, B and C. (fig. 1). Site A is the point of entry of Samaru stream into the lake. Site B is at the intake structure for the water purification plant at A.B.U. main campus, and site C is midway between the dam and site B. Table 4 includes the mean 10-month densities of faecal coliforms and faecal streptococci at sites B and C. Mean 10-month faecal coliform density at B was 2.2 x 10^2/100ml while at C, it was 8.2 x 10^2/100ml. For faecal streptococci, the 10-month mean for B was 1 x 10^1/100ml and for C, 1.1 x 10^2/100ml. Over the 41-week sampling period, there were only 2 (4%) positive faecal coliform samples out of 41 samples taken at B. At C, 4 (9%) samples were positive for faecal coliforms. Site B showed 4 (9%) samples positive out of 41 samples analysed for faecal streptococci; while at C, 6 (19%) samples, were positive (see appendices A and B).

TABLE 3

MEAN DRY SEASON DENSITIES FOR FC AND FS

AT SITES 5 - 9 AND A

SITES	FAECAL COLIFORMS/100ml	FAECAL STREPT./100ml
5	5.5×10^3	1.5×10^3
6	2.6×10^4	1.5×10^3
7	2.75×10^3	4.3×10^3
8	5.1×10^3	1×10^3
9	2.3×10^3	1×10^3
A	1×10^2	1×10^1

FC = Faecal Coliforms

FS = Faecal Streptococci

DRY SEASON = October - March

IBRAHIM

Table 4: Mean Densities of faecal coliforms and faecal streptococci at site B and site C on Kubanni Lake.

Site	Indicator	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.
	F C	-	-	-	-	-	-	$2.5x10^1$	-		$5x10^2$
B	F S	-	-	$2.5x10^4$						$2.5x10^1$	$2.5x10^1$
	F C	$2.5x10^3$		-	-	$2.5x10^2$	-	$2.5x10^1$	-	-	$2.5x10^2$
C	F S		$2.5x10^4$	$2.5x10^3$	$5x10^3$	-	-	-	-	$7.5x10^1$	-

Site B over intake structure for the purification plant

Site C midway between dam wall and spillway

F. C. faecal coliforms/100 ml.

F. S. faecal streptococci/100 ml.

The mean 10-month faecal coliform and faecal streptococcus densities at sites B and C are presented in Table 4. The table illustrates the mean counts for each of the 10-months (April, 1977 - January, 1978) during which samples were collected.

The positive faecal coliform samples for site B occurred in October and January. For site C, they were in April, August, October, and January. The positive faecal streptococcus samples at B were obtained in June, December and January while at site C, they were obtained in May, June, July abd December. About twice as many positive samples were obtained at site C as at B (table 4). Incidence of faecal streptococci appeared to be higher than of coliforms in the lake.

The mean dry season (October - December, 1977) counts for site A is given in table 3. Faecal coliform density for the three-month period was 1 x 10^2/100ml while faecal streptococcus density for the same period was 1 x 10^1/100ml.

ISOLATION OF SALMONELLA

Little success was achieved in the attempts to isolate Salmonellae from all the sites. Although all samples were screened for Salmonellae only five were isolated and identified at sites 1, 2 and 3, by biochemical tests. Two of these were sent to a reference laboratory for serological identification.

DISCUSSION

The efficiency of the sewage treatment plant at A.B.U. main campus (fig. 6) was measured. The efficiency is reported as percentage removal of faecal coliforms from site 1 to site 2. There was a variation in ability of the plant to reduce faecal coliform numbers between April 1977 and January 1978 (Table a). Only in April 1977 did the plant appear to perform with considerable efficiency. This period coincides with a time when students were away on Easter vacation. It's efficiency was reasonably high during May (91%) and August (96%). The high efficiency in May can be explained by the fact that the Easter vacation extended to the first week of that month. August efficiency was that high probably because of the long vacation. At all other times in the sampling period, the plant's efficiency was only moderate.

From the visual observation of the turbidity of the plant effluents (fig. 7), it would appear that a high concentration of organic matter is released from the plant, with effluents. Therefore, faecal coliforms and faecal streptococci may tend to grow in the effluents. This statement is justifiable by the high counts of both organisms observed for sites 2 and 3. Even in the rainy season when dilution of effluents was expected to be high (appendix E), there appeared to be no reduction in numbers. This may have been due to run-off from Area 'A' gully and from the surroundings of the effluent gully. As a result of the above observations, it may be concluded that the sewage treatment plant did not function satisfactorily most of the time especially when students were on - campus.



Fig. 6 A.B.U. Sewage Treatment Plant (Site 1)



Fig.7 Outlet from A.B.U.Sewage Treatment Plant (Site 2)

The situation was improved slightly during the period Mid - April - May ending 1978, when all students were off - campus due to students unrest. There was a progressive decrease in faecal coliform counts from sites 2 - 6 with the organism disappearing completely from sites 7, 8, and 9. Similarly, faecal streptococci disappeared from sites 3 - 9 during the same period (Table 2).

It was also observed that faecal coliform and faecal streptococcal counts are high during the dry season (figs. 3 and 4). Normally, one would expect some reduction in numbers to take place due to sedimentation, aeration and filteration. It would therefore appear that the organic matter in effluents was sufficient for the bacteria to maintain a steady population between sites 2 and 3, 600m long.

The drop in counts observed in August (fig. 3) for the four sites is attributable to the long vacation and the fact that dilution was highest in that month. The highest rainfall of 302mm in 1977 occured in August 1977 (appendix E). The counts increased gradually from September onwards as students gradually returned. The slight drop in counts at sites 1 - 4 (fig. 3) in December 1977 and January 1978 may be due to absence of students from the campus for the christmas vacation.



Fig. 8a Farmer drawing water from effluent gully.



Fig. 8b Farmer irrigating crops with effluent

NOTE:

8(b) Farmer is pouring effluents directly on tomatoes.

at the farms irrigated with A.B.U. sewage effluents. It is also observed that the stretch of stream below Jama'a village is used by livestock (cattle), for grazing. As the stream is contaminated by the sewage effluents, it is highly possible that the cattle be infected by Salmonella. Therefore, the possibility of zoonotic transfers between cattle and man cannot be ruled out. Olayemi et al.(1978) isolated 13 different serotypes of Salmonellae from cattle slaughtered at Zaria abattoir. Two of these have been isolated from patients suffering gastroenteritis in Nigeria (Ojo, 1972). In this study, only five Salmonellae were isolated from effluents,and were identified biochemically. However, higher recoveries from the sewage effluents were expected considering the high FC counts in the sewage effluents (Kampelmacher and van Noorle Jensen 1976; and Dukta and Bell 1973). The low recovery rate observed could have been due to the low survival rate of Salmonellae in water and the possible side effect of the selective media used for isolation, on the growth of Salmonella cells. (Hoadley and Cheng 1973).

The interelationship between Salmonellae isolated from cattle, man and sewage effluents has not been determined. Further work in this respect is highly desirable in order to establish any possible relationship.

From the present study, the necessity to improve the sewage effluents before discharge into the Kubanni River becomes urgent.

Ogunrombi (1974), suggested chlorination of the effluent before discharge into the Kubanni River; but chlorination alone may not be sufficient because faecal bacteria are known to grow in chlorinated effluents depending on the concentration of residual chlorine. From the studies of Silvey at al. (1974); Davis and Keen (1974); Milbauer and Grosswicz (1959); Shuval et al. (1972); Braswell and Hoadley (1974); and Lin, (1976); it is shown that faecal bacteria can regrow after chlorination, in effluents. If the effluents are to be used for irrigation, groundwater recharge and augmentation of flow in Kubanni River (Ogunrombi. 1974), then a more effective secondary treatment of effluents, such as soil filteration is to be recommended. Van Donsel (1964); Pound and Crites (1973), and Edmonds, (1976); all support soil filteration of sewage effluents. Alternatively, secondary treatment by maturation ponds may be an advantage. Tables 1,3, and 4 show the FC and FS densities in and around the Kubanni Lake. From these results, it is observed that the level of contamination in the lake is low at this time when the lake is static. However, the level of contamination is expected to rise when the lake is in use. It is believed that during pumping of the water into the purification plant, the sediments at the bottom of the lake may be agitated and therefore, some of the pollutants from the sediments may be introduced into the water layer above. The above statement is

justified by the work of van Donsel and Geldreich (1971). They found that there was a 100 - 1000 fold increase in coliform counts in bottom mud than in the over - lying water. They could also isolate more Salmonella from bottom mud than from the overlying water.

The bacterial counts from Samaru Stream as measured at site A was low (Table 2), compared with observed counts of Smith and Gaultney. (personal communication) The observed low faecal indicator densities in this study was recorded during the dry season when there was no flow in the stream; whereas the above workers took measurements in the rainy season when pollutants are expected to be washed into the lake.

Generally, low densities of faecal coliforms and faecal streptococci were recorded for the lake (Tables 1,3, and 4). That no Salmonella was isolated from lake water in this study should not give a false sense of security. The observed FC and FS contamination in the lake are probably of animal origin but human contamination cannot be ruled out.

Finally, the disposal of the [illegible]

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FECAL COLIFORM COUNTS/100MLS

Weeks	1	2	3	4	A	B
1	$5.8x10^6$	$1x10^6$	$5x10^7$	ND	-	-
2	$1.9x10^8$	$1x10^6$	$1x10^7$	"	-	-
3	$1x10^8$	TD	TD	"	-	-
4	$3x10^7$	$2x10^6$	$1.3x10^6$	"	-	$5x10^3$
5	$8x10^7$	$2x10^6$	$4x10^5$	"	-	-
6	$1x10^7$	$3x10^6$	$4x10^6$	"	-	-
7	$2x10^7$	$3x10^6$	$1x10^6$	"	-	-
8	$5x10^6$	$2x10^6$	$2x10^7$	"	-	-
9	$1x10^5$	$4x10^6$	$1x10^5$	"	-	-
10	$3x10^6$	$4x10^6$	$2x10^7$	"	-	-
11	$4x10^7$	$1x10^6$	$3x10^7$	$1x10^5$	-	-
12	$7x10^6$	$3x10^6$	$1x10^7$	$2x10^5$	-	-
13	$5x10^7$	$6x10^6$	$3x10^6$	$3x10^5$	-	-
14	$1x10^7$	$5x10^6$	$1.5x10^6$	$1x10^6$	-	-
15	$1x10^7$	$3x10^6$	$3x10^5$	$6x10^5$	-	-
16	$7x10^5$	$1x10^6$	$1x10^5$	$1x10^5$	-	-
17	$5x10^6$	$1x10^5$	$2x10^5$	$5x10^3$	-	$1x10^3$
18	$1x10^7$	$1x10^6$	$2x10^5$	$1x10^4$	-	-
19	$5x10^6$	$1x10^6$	$1x10^5$	$2x10^5$	-	-
20	$4x10^6$	$2x10^6$	$1.4x10^6$	$2x10^5$	-	-
21	$8x10^6$	$1x10^6$	$3x10^6$	$5x10^4$	-	-
22	$1x10^7$	$3x10^6$	$3x10^6$	$3x10^6$	-	-
23	$1x10^7$	$2x10^6$	$1x10^6$	$2x10^6$	-	-
24	$3x10^6$	$3x10^6$	$3x10^6$	$3x10^6$	-	-
25	$1x10^7$	$2x10^7$	$3x10^6$	$3x10^6$	$1x10^2$	

Weeks	1	2	3	4	A	B
26	$2x10^7$	$2x10^7$	$2x10^6$	$2x10^6$	-	$1x10x^2$
27	$1x10^6$	$1x10^6$	$1x10^5$	$2x10^6$	-	-
28	$1x10^7$	$1x10^7$	$4x10^6$	$2x10^5$	-	-
29	$1x10^7$	$2x10^6$	$2x10^6$	$2x10^6$	-	-
30	$4x10^7$	$1x10^7$	$5x10^6$	$2x10^6$	-	-
31	$7x10^7$	$6x10^7$	$4x10^6$	$2x10^6$	-	-
32	$5x10^7$	$4x10^6$	$4x10^6$	$1x10^6$	-	-
33	$2x10^7$	$9x10^6$	$1x10^6$	$1x10^6$	-	-
34	$4x10^7$	$1x10^7$	$1x10^6$	$1x10^6$	-	-
35	$6x10^7$	$7x10^7$	$2x10^6$	$1x10^6$	-	-
36	$8x10^7$	$1x10^7$	$3x10^6$	$1x10^6$	-	-
37	$3x10^7$	$1x10^7$	$1x10^6$	$1x10^6$	-	-
38	$6x10^7$	$5x10^5$	$7x10^5$	$1x10^6$	$2x10^3$	$1x10^3$
39	$3x10^7$	$2x10^7$	$1x10^6$	$1x10^6$	-	-
40	$4x10^7$	$3x10^6$	$1x10^6$	$8x10^5$	-	-
41	$2x10^7$	$3x10^7$	$2x10^6$	$2x10^6$	-	-

Fecal Streptococci Counts/100MLS

Weeks	1	2	3	4	A	B
1	TD	TD	TD	ND	-	-
2	"	"	"	"	-	-
3	"	"	"	"	-	-
4	$1.6x10^6$	$1x10^5$	$1x10^5$	"	-	-
5	$3x10^7$	TD	TD	"	-	-
6	$1.7x10^6$	$2x10^6$	$1x10^6$	"	-	$1x10^5$
7	$2x10^7$	$2x10^6$	$1x10^7$	"	-	-
8	$4x10^6$	$2x10^6$	$1x10^6$	"	-	-
9	$6x10^6$	$1x10^7$	$4x10^6$	"	$1x10^6$	$1x10^4$
10	$1x10^6$	ND	ND	"	-	-
11	$8x10^6$	"	"	"	-	-
12	$7x10^7$	$1x10^7$	$4x10^5$	$2x10^5$	-	-
13	$8x10^6$	$3x10^5$	TD	$1x10^5$	-	-
14	$1x10^7$	$4x10^5$	$3x10^5$	$1x10^6$	-	-
15	$4x10^7$	$6x10^5$	$1x10^5$	$8x10^4$	-	$2x10^4$
16	$1x10^6$	$3x10^5$	TD	$4x10^1$	-	-
17	$3x10^7$	$3x10^5$	$1x10^5$	-	-	-
18	$1x10^7$	$2x10^5$	$1x10^5$	-	-	-
19						
20						
21	NO	M E D I	A			
22						
23						
24						
25						
26	$2x10^6$	$2x10^6$	TD	$2x10^5$	-	-
27	$1x10^7$	$2x10^5$	"	$2x10^5$	-	-

Weeks	1	2	3	4	A	B
28	$6x10^6$	$3x10^6$	$1x10^6$	$5x10^5$	-	-
29	$5x10^6$	$8x10^6$	-	$5x10^5$	-	-
30	$1x10^7$	$1x10^7$	$2x10^6$	$2x10^6$	-	-
31	$6x10^7$	$5x10^6$	$1x10^6$	$2x10^5$	-	-
32	$2x10^7$	$6x10^6$	$2x10^6$	$2x10^6$	-	-
33	$2x10^7$	$2x10^6$	$1x10^6$	$1x10^6$	$1x10^2$	$1x10^2$
34	$3x10^5$	$3x10^6$	$1x10^5$	$4x10^5$	-	$1x10^2$
35	$4x10^6$	$5x10^6$	$2x10^6$	$4x10^5$	-	$1x10^2$
36	$5x10^6$	$3x10^6$	$3x10^6$	$3x10^5$	-	-
37	$8x10^6$	$1x10^6$	$2x10^5$	$2x10^5$	$1x10^2$	-
38	$5x10^7$	$3x10^5$	$3x10^5$	$1x10^6$	-	-
39	$6x10^6$	$1x10^6$	$3x10^5$	$3x10^5$	-	-
40	$9x10^6$	$1x10^6$	$1x10^5$	$2x10^5$	-	-
41	$5x10^6$	$1x10^6$	$3x10^5$	$2x10^5$	-	-

APPENDIX C

Lactose Positive Counts/100MLS

Weeks	1	2	3	4	A	B
1	$1x10^8$	$3.1x10^7$	$4.2x10^6$	ND	-	$1x10^5$
2	$5x10^7$	$4.3x10^7$	$1x10^5$	"	-	-
3	$1.6x10^6$	TD	TD	"	-	-
4	$3x10^8$	$1.2x10^7$	$3x10^7$	"	$6x10^3$	$8x10^3$
5	$1x10^8$	TD	TD	"	-	-

Weeks	1	2	3	4	A	B
6	$4x10^7$	$1.4x10^6$	$1.7x10^7$	ND	-	$1x10^5$
7	$5x10^7$	$8x10^6$	$2x10^6$	"	$1x10^4$	$4x10^3$
8	$1.4x10^7$	$8x10^6$	$8x10^6$	"	-	-
9	$1x10^8$	$1x10^6$	$2x10^6$	"	-	-
10	$1x10^7$	ND	ND	"	-	$1x10^4$
11	ND	"	"	"	-	-
12	$1x10^8$	$3x10^5$	$6x10^6$	$5x10^5$	-	-
13	$5x10^5$	$1x10^6$	TD	$8x10^5$	-	-
14	$2x10^7$	$3x10^6$	$5x10^5$	$5x10^6$	-	$2x10^3$
15	$2x10^7$	$3x10^3$	$1x10^6$	$4x10^5$	-	$4x10^3$
16	$8x10^7$	$1x10^7$	$4x10^6$	$3x10^5$	-	-
17	$5x10^6$	$5x10^6$	$2x10^6$	$5x10^4$	$2x10^4$	$1x10^4$
18	$9x10^6$	$2x10^6$	-	$4x10^3$	-	-
19	$8x10^6$	$2x10^6$	TD	$1x10^5$	-	-
20	$1.5x10^7$	$3x10^6$	$1x10^6$	$3x10^5$	-	-
21	$2x10^7$	$4x10^6$	$5x10^6$	$6x10^4$	-	-
22	$5x10^7$	$5x10^6$	$6x10^6$	$4x10^6$	-	$3x10^2$
23	$4x10^7$	$3x10^6$	$3x10^6$	$2x10^6$	-	$1x10^3$
24	$4x10^6$	$5x10^6$	$5x10^6$	$3x10^6$	-	$1x10^2$
25	$5x10^7$	$2x10^7$	$6x10^6$	$5x10^6$	-	-
26	$1x10^7$	$3x10^7$	$1x10^6$	$2x10^6$	-	$1x10^2$
27	$5x10^6$	$3x10^6$	$1x10^6$	$3x10^6$	-	-
28	$7x10^7$	$1x10^7$	$5x10^6$	$1x10^5$	$1x10^3$	-
29	$3x10^7$	$3x10^7$	$3x10^6$	$3x10^6$	-	-
30	$1.5x10^7$	TD	TD	$1x10^6$	-	$2x10^2$
31	$4x10^7$	$4x10^7$	$3x10^6$	$3x10^5$	-	-
32	$4x10^7$	$7x10^6$	$3x10^6$	$2x10^6$	-	$1x10^2$
33	$5x10^7$	$2x10^7$	$1x10^6$	$1x10^6$	$6x10^2$	$1x10^3$

Weeks	1	2	3	4	A	B
34	$6x10^7$	$1x10^7$	$2x10^6$	$6x10^5$	$3x10^2$	$1x10^3$
35	$2x10^7$	$2x10^7$	$x10^6$	$1x10^6$	$3x10^2$	$2x10^2$
36	$2x10^7$	$1x10^7$	$6x10^6$	$5x10^5$	$4x10^2$	$2x10^2$
37	$4x10^7$	$7x10^6$	$2x10^6$	$3x10^5$	$4x10^2$	$1x10^3$
38	$4x10^7$	$1x10^6$	$1x10^6$	$2x10^6$	-	-
39	$4x10^7$	$2x10^7$	$2x10^6$	$1x10^6$	$1x10^2$	$1x10^2$
40	$4x10^7$	$1x10^7$	$3x10^6$	$1.3x10^5$	$1x10^2$	$1x10^2$
41	$2x10^7$	$x10^2$	$2x10^6$	$1x10^6$	-	-

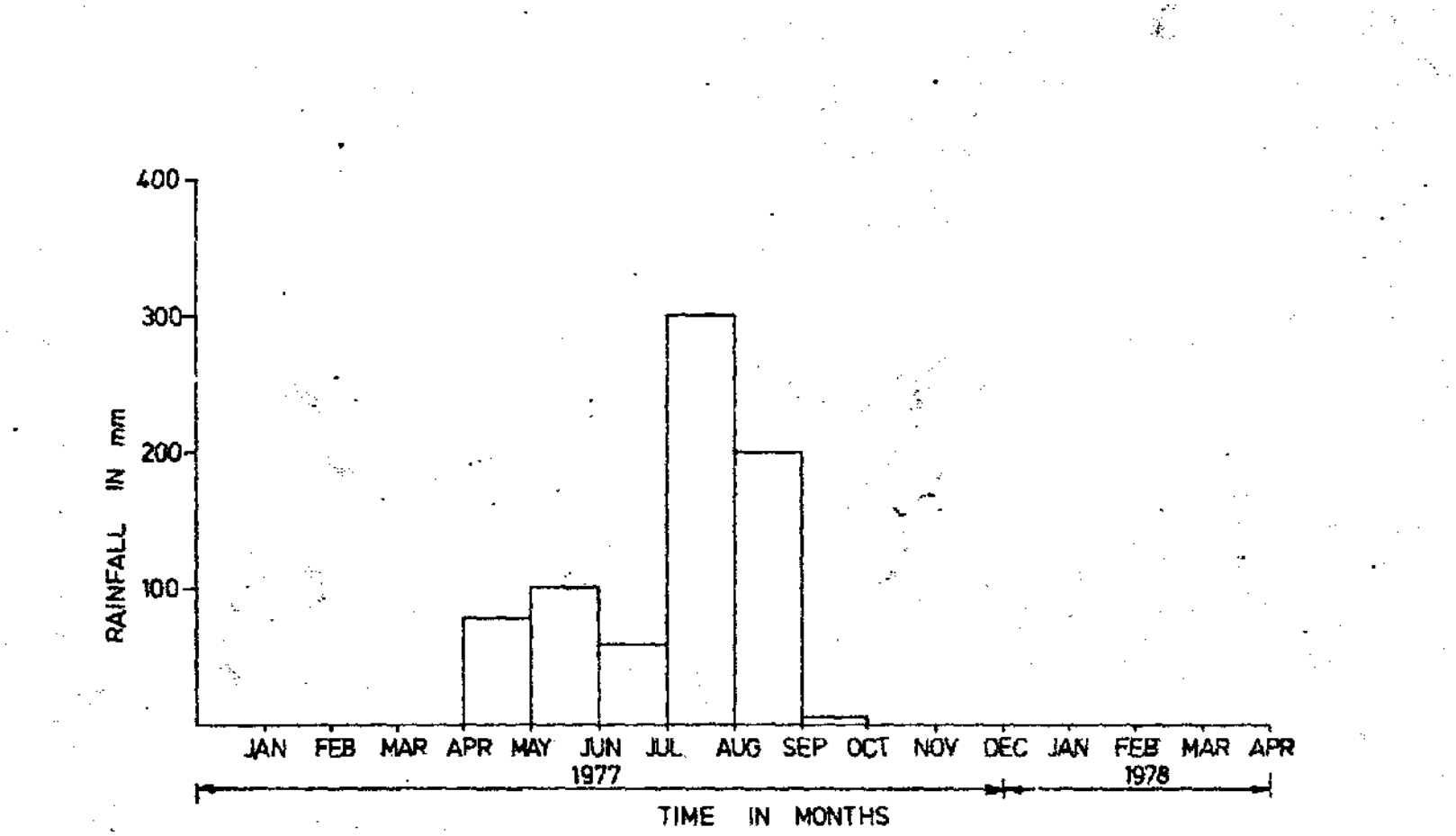
APPENDIX D

Lactose Negative Counts/100MLS

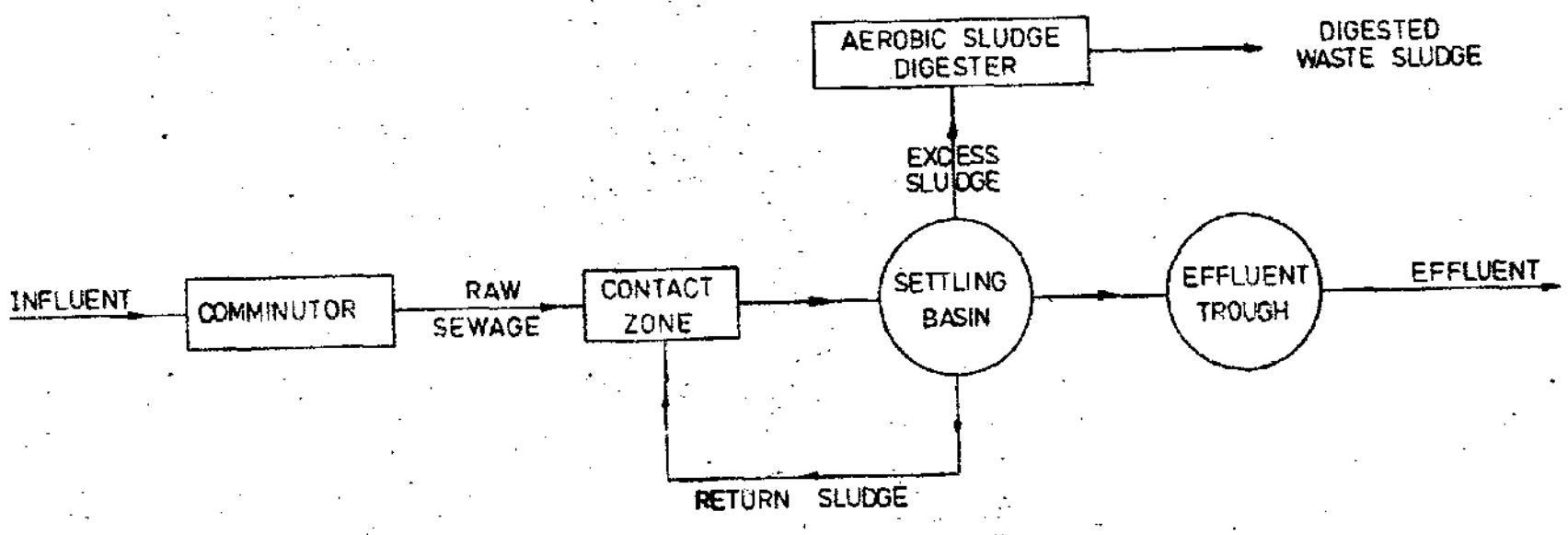
Weeks	1	2	3	4	A	B
1	$3x10^8$	$6x10^7$	$1.1x10^6$	ND	-	$1x10^5$
2	$1.9x10^8$	$4x10^7$	$3x10^7$	"	-	-
3		ND	ND	"	-	-
4	$4x10^8$	$1.2x10^7$	$4x10^6$	"	$5x10^3$	$8x10^3$
5	$3x10^8$	TD	TD	"	-	-
6	$2x10^7$	$4x10^6$	$1.1x10^7$	"	-	$4x10^3$

Weeks	1	2	3	4	A	B
7	$2x10^7$	$3x10^6$	$7x10^6$	ND	$5x10^4$	$8x10^4$
8	$7x10^6$	$7x10^6$	$1x10^7$	"	$1x10^3$	$1x10^3$
9	$2x10^8$	$2x10^6$	$1x10^6$	"	-	$3x10^6$
10	$1x10^7$	ND	ND	"	-	$2x10^3$
11	TD	"	"	"	-	-
12	$4x10^7$	$3x10^6$	$1.5x10^6$	$3x10^5$	-	-
13	$1x10^8$	$1x10^6$	$1x10^5$	$1x10^6$	-	-
14	$1x10^8$	$8x10^5$	$1.2x10^6$	$3x10^6$	$2x10^3$	$1x10^4$
15	$2x10^7$	$5x10^6$	$3x10^3$	$1.5x10^6$	$1x10^4$	$1x10^3$
16	$4x10^7$	$1x10^5$	$3x10^4$	$2x10^5$	-	-
17	$4x10^6$	TD	$1x10^6$	$1x10^5$	$1x10^4$	$2x10^4$
18	$1x10^7$	$8x10^6$	$6x10^5$	$2x10^4$	$8x10^3$	$1x10^3$
19	$8x10^6$	$3x10^6$	$3x10^6$	$3x10^5$	-	$1x10^3$
20	$6x10^6$	$2x10^6$	$4x10^6$	$2x10^5$	-	-
21	$3x10^7$	$7x10^6$	$4x10^6$	$1.7x10^5$	-	$1x10^3$
22	$5x10^7$	$2x10^6$	$7x10^6$	$5x10^6$	-	-
23	$5x10^6$	$2x10^6$	$2.5x10^6$	$1.2x10^6$	-	-
24	$4x10^6$	$4x10^6$	$5x10^6$	$5x10^6$	-	-
25	$3x10^7$	$2x10^7$	$4x10^6$	$4x10^6$	$1x10^3$	-
26	$1x10^7$	$7x10^6$	$3x10^6$	$1x10^6$	-	-
27	$3x10^5$	$4x10^5$	$2x10^5$	$2x10^5$	-	-
28	$5x10^6$	$1x10^7$	$1x10^6$	$4x10^4$	-	$2x10^2$
29	$1x10^6$	$2x10^6$	TD	$1x10^6$	-	-
30	$2x10^7$	TD	TD	$7x10^5$	$1x10^2$	-
31	$4x10^7$	$1x10^7$	$3x10^6$		-	-

Weeks	1	2	3	4	A	B
32	$3x10^7$	$1x10^7$	$1x10^6$	$1x10^6$	-	-
33	$3x10^7$	$1x10^7$	$2x10^6$	$1x10^6$	-	-
34	$4x10^7$	$5x10^6$	-	$1x10^6$	-	$2x10^2$
35	$9x10^6$	$1x10^7$	$1x10^6$	$3x10^5$	$1x10^2$	$2x10^2$
36	$4x10^7$	$8x10^6$	$1x10^6$	$4x10^5$	$1x10^2$	$2x10^2$
37	$5x10^7$	$3x10^6$	$2x10^6$	$1x10^5$	$4x10^2$	$1x10^3$
38	$5x10^7$	$3x10^5$	$1x10^6$	$1x10^6$	-	-
39	$1x10^7$	$1x10^6$	-	$4x10^5$	-	-
40	$1x10^7$	$2x10^6$	$3x10^5$	$4x10^5$	-	-
41	$3x10^6$	$1x10^7$	$1x10^6$	-	-	-



APPENDIX E : MEAN MONTHLY RAINFALL IN ZARIA. (JAN. 1977 - APR 1978)



APPENDIX F : FLOW DIAGRAM OF STEPS IN SEWAGE TREATMENT BY OXIGEST PACKAGE PLANT MODEL 'R'.